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Full length Research Article

Gestational Administration of Aqueous Leaf Extract of *Jatropha tanjorensis* Alleviates Postpartum-like Behaviours in Experimental Rats

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Summary: This study examines the effect of gestational administration of aqueous leaf extract of *Jatropha tanjorensis* (JT) on postpartum-like behavioural outcomes to delineate its possibility as a prophylactic, therapeutic agent in the treatment of postpartum symptoms. Inseminated female rats (120-150g) were grouped into two-control and JT group (n=10). Control received 20 ml/kg of distilled water and JT group received 500 mg/kg of JT orally once daily for 21 days in gestation. Non-pregnant rats were excluded from the study. Parameters assessed at postpartum include antidepressant-like (force swim test, FST; tail suspension-test, TST), locomotor (open field test, OFT), anxiolytic-like (elevated plus maze, EPM; light-dark box, LDB), learning and memory (T-maze; novel object recognition task, NORT), social (nest score) and analgesic-like (hot plate test, HPT; tail flick test, TFT) behaviours. JT increased ($P<0.05$) mobility and latency to immobility durations in FST and TST; open arm entry ($P<0.001$) and duration ($P<0.01$) in EPM and light box duration ($P<0.05$) in LDB; locomotion and exploration, but reduced anxiety-like levels in EPM, LDB and OFT. It increased nest score ($P<0.05$); mean retraction time ($P<0.01$) of TFT. JT showed positive score for short and long term memory in NORT and improved percentage alternation in T-maze though not significant compared to control. In conclusion, the aqueous extract has a therapeutic effect that reduces postpartum-like depression and anxiety, and improves locomotor activity. JT can be a preventive and adjuvant therapeutic option for pregnant women.

Keywords*: Jatropha, postpartum disorder, Emotionality, Pain, Gestation*

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INTRODUCTION

In females, pregnancy, delivery, and postpartum period are uniquely characterized by extremely varying behavioural changes. However, in a large subset of mothers, these phases also represent a risk period for the development of postpartum disorders such as postpartum depression (PPD), anxiety (PPA) and cognitive deficits (Zhang *et al*., 2021). PPD and PPA have many negative consequences on maternal health (Ali *et al*., 2013), which include biological and psychological factors, risky behaviours and mother-child interactions (Slomian *et al*., 2019; Field, 2017). Cognitive decline was also reported to have consequence on mother-child interactions (Postma *et al*., 2014; Albin-Brooks *et al*., 2017). It is estimated that mothers display about 10-20% PPA and over 25% PPD (Cárdenas *et al*., 2020), and over 80% cognitive decline (Qiu *et al*., 2021). The main predictor for postpartum disorders is an antenatal episode of the illness (Grant *et al*., 2008; Topiwala *et al*., 2012). However, postpartum hyperalgesia due to acute pain after delivery was shown to increase the development of postpartum disorder (Eisenach *et al*., 2008; Lim *et al*., 2018). Evidence from animal and clinical studies indicated that pain sensation could change from acute pain caused by tissue damage to a complex and multifaceted pain syndrome (Maldonado and De Jesus, 2021; Quesada *et al*., 2021). However, these symptoms in postpartum women are often overlooked, leading to lower diagnosis rates and even lower treatment rates (Zhang *et al*., 2021).

Jatropha tanjorensis is a perennial herb normally grown in Southern Nigeria commonly called 'Hospital too far' (Oyewole *et al*., 2012). Phytochemical screening of JT leaf revealed that it contains bioactive constituents such as alkaloids, flavonoids, tannins, cardiac glycosides, anthraquinones and saponins (Iroanya *et al*., 2018). Cook and Samman, (1996) reported that flavonoids due to their high antioxidant capacity have health-promoting properties and help reduce the risk of diseases. Bribi *et al*., (2017) and Contreras *et al*., (2017) in their separate studies observed that alkaloid exert antioxidant and analgesic activities, while Feng *et al*., (2006) reported anti-inflammatory and anxiolytic activities of plant alkaloids. Albuquerque *et al*., (2005) reported that tannins are known to heal wounds and inflamed mucous membranes, while Wagner and Elmadfa, (2003) reported that they possesses antioxidant properties. Madubuike, *et al*., (2015) reported that the leaf extract of JT

possesses antioxidant and anti-diabetic properties. Iroanya *et al*., (2018) reported that JT improve haematological indices which revealed an enhancement of bone marrow function. Falodun *et al*., (2013) reported that JT increases the amount of iron available for erythropoiesis, increase packed cell volume and hemoglobin concentration in rats and recommended the extracts usage in physiological conditions like pregnancy and during menstruation. Nindaratnasari, (2017) reported JT usage to ease baby delivering process.

Despite the well documented multi-dimensional usage of *Jatropha tanjorensis*, there is no report on the behavioural effects of the plant extract on maternal health. Hence, this study describes the effect of gestational administration of an aqueous leaf extract from JT on several postpartum-like behavioural outcomes to delineate the possibility of using JT as a prophylactic, therapeutic agent in the treatment of postpartum symptoms. This paper could be the first or one of the first few studies examining the effect of JT on neurological aspects of postpartum responses in rats. One of the strengths of this paper is the extensive behavioural assessments showing several measures of neuropsychiatric, social, nociceptive, and cognitive responses. However, the paper lacks important underlying molecular explanations of the behavioural outcomes. A look at HPA/HPG/oxytocin levels might improve the robustness of the data.

MATERIALS AND METHODS

Preparation of Extract: The leaves of *Jatropha tanjorensis* were harvested from the botanical garden of the University of Calabar, Nigeria. A voucher specimen has been kept in the botanical garden (Herb/Bot/UCC/182). They were prepared as described by Agarwal *et al*., (2007). The leaves was thoroughly washed with clean water and air-dried at room temperature for two days, then further dried in an oven at 40°C for 24 hours. The crispy leaves were then ground into fine powder and preserved in moisture-free, airtight laboratory containers for further use. The powdered plant material (100 g) was macerated with water (1000 ml) in ratio of 1g of the powdered plant to 10ml of water and was agitated intermittently for 48 hours, filtered into a clean glass jar. Filtration was done using a Whatman No 1 filter paper to separate the filtrate from the residue. The filter paper was folded into four portions and put in the funnel and placed into 1,000ml beaker, the filtrate containing the extract was carefully poured into the funnel which filtered into the beaker through the filter paper. After obtaining the filtrate, it was then poured into an evaporating dish which was thereafter dried on a thermostatically controlled water bath at 42 0C. The drying was monitored until it turned into a paste form. The yield of the aqueous extract of JT weighed 73g. The extraction method closely represents how the leaves might be consumed locally. Hence the results have a tendency for a more direct application.

Determination of LD_{50}: The LD_{50} was determined by the method of Lorke, (1983) using 12 female albino Wistar rats which comprised of two phases. In the first phase the rats were divided into 3 groups of 3 rats each and were treated with the aqueous leaf extract of JT at doses of 10, 100 and1000mg/kg body weight orally. In the second phase 3 rats were divided into 3 groups of 1 rat each and were treated with the aqueous leaf extract of JT at doses of 1000, 1600, 2900, and 5000mg/kg body weight orally. In both phases the animals were observed for 24h for signs of toxicity as well as mortality. There was no toxicity and mortality recorded even at the highest dose of 5000mg/kg. Therefore, in this study the extract dosage of 500mg/kg was considered safe for pregnant rats.

Experimental animals: Thirty albino rats (20 females and 10 males) weighing between 120 -150g were obtained from the College of Medical Sciences animal house of University Calabar, Cross River State, Nigeria. The animals were kept under standard laboratory conditions and housed in well ventilated plastic cages at room temperature and relative humidity with light and dark cycles (12hr/12hr). The animals were acclimatized for one week and were provided standard rat pellet (Pfizer feed PLC, Lagos, Nigeria) and water ad libitum.

Ethical consideration: Ethical approval was obtained from the University of Calabar College Ethical Committee on the use of experimental animals with protocol number (093PHY3321).

Experimental design: In the model of natural pregnancy, rats were caged at a ratio of 1 male to 2 females, and the appearance of vaginal plug was considered day 1 of gestation (Zhang *et al*., 2021). The inseminated female rats were randomly assigned into two groups. Group 1 served as control (n=10) which received 20 ml/kg of distilled water (vehicle) orally, while Group 2 served as JT treated (n=10) and received 500mg/kg of JT orally for 21 days throughout gestation. After two weeks of observation, the number of inseminated female rat not pregnant rats was excluded from the study. The pregnant rats for control (n=7) and for JT (n=9).

Neurobehavioural assessments of postpartum rats:

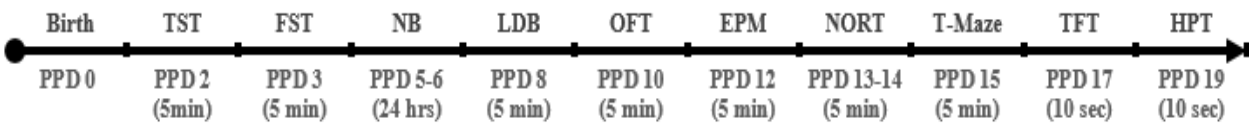

Plate 1:
Experimental design for neurobehavioural studies and durations. Tail suspension test (TST), Force swim test (FST), Nesting behaviour (NB), Light dark box (LDB), Open field test (OFT), Elevated plus maze (EPM), Novel object recognition task (NORT), Tail flick test (TFT), Hot plate test (HPT), Postpartum day (PPD)

Assessment for anti-depressant activity

Force swim test: A transparent container of diameter 15 cm, height 25 cm and with a water depth of 20 cm was allowed to sit overnight to maintain a temperature at 30°C ± 1°C. At this depth of water, rat was unable to touch the bottom or the side walls of the container with their paws or tail. On PPD 3 each dam was allowed to swim in the transparent container for 5 min and the behavioural score was recorded using a stopwatch. After the test session, the dam was scooped up from the transparent container with a plastic container and placed in a holding cage filled with paper towel to dry before being returned to their home cages. After each trial, the water was changed. Behaviour scored include latency to immobility, mobility, immobility and frequency of defecation (Englisch *et al*., 2009).

Tail suspension test (TST): A tail suspension box made of wood with dimensions (55 height x 60 width x 11.5 cm depth) was used. The compartment width and depth was sufficiently sized so that the dam couldn't make contact with the walls. On PPD 2, each dam was suspended by hanging by the tail using adhesive tape and its body hung down in the air. TST is based on the assumption that the animal will try to escape the stressful situation. After a certain time, the animal ceases to struggle and immobility occurs; longer immobility phases are sign of depressive behaviour. Test was carried out in a lit room with minimal background noise for a period of 5 min. Behavioural Score include mobility, immobility and latency to immobility (Englisch *et al*., 2009).

Assessment of locomotion and emotionality

The open field test: The OFT apparatus is a white painted plywood (72 X 72 cm) with blue lines drawn on the floor to divide it into 18 x 18 cm squares. A central square (18 x 18 cm) is drawn in the middle of the open field. On PPD 10 each dam was placed in the center of the apparatus and allowed to explore the area for 5 min. Behaviours Scored include line crossing, center square entry (CSE), rearing, stretch attend postures (SAP), grooming and freezing. Each dam was then given a score for total locomotor activity and emotionality that was calculated as the sum of line crosses and number of rears (Bisong *et al*., 2010). The apparatus was cleaned with 70 % ethanol after each trial.

Assessment of anti-anxiety activity

Elevated plus-maze test: The EPM apparatus consists of two open arms (30 × 5 cm), and two closed arms (30 × 5 × 15 cm), elevated 25 cm above floor level. The arms are connected to a central square (5 × 5cm) where the dam is placed. The edges, 4 mm high, surrounds the open arms, reducing the chances of rats falling from the apparatus. On PPD 12, each dam was individually placed in the center square of the apparatus and allowed to freely explore the apparatus for 5 min. The dam behavior was recorded for the test period of 5 min and then analyzed. After each dam, the entire maze was cleaned with 70% ethanol. Behaviour scored include, open arm entry, closed arm entry, head dips, SAP, grooming, rearing, urination and defecation (Sutulovic *et al*., 2021).

Light/dark transition box: The apparatus is made of a rectangular box (45 x 27 x 27 cm), partitioned into two unequal compartments. The box has two compartments, a light and a dark one. The dark chamber is smaller and often is the compartment considered as safe by the mice. The small compartment (18 x 27 cm) was painted black and the larger compartment (27 x 27 cm) was painted white. These compartments were connected by a door (7.5 x 7.5 cm) located at floor level in the center of the wall between the two compartments. The floor was divided into 9 x 9 cm squares and covered with plexiglas. Both compartments were covered with lids of clear plexiglas. A 60-watt table lamp located 40 cm above the center of the white compartment provided bright illumination of white light. On PPD 8, each dam was placed in the center of the white compartment facing the door and allowed to explore the apparatus for 5 min. The dam's behaviour was recorded by a digital camera set up high above the plexiglass. After each dam, the entire apparatus was cleaned with 70% ethanol. Behaviours scored include transitions, line crosses, rearing, SAP, grooming, dark box and light box duration (Bisong *et al*., 2017).

Assessment of learning and memory

Novel object recognition test: The NORT assesses rodent's ability to recognize a familiar object over a variable length of time; this ability has been coined recognition memory (Ajiwhen and Bisong, 2013). The NORT modified by Shimoda *et al*., (2021) was used to test cognitive memory. On PPD 13, there is an initial habituation to the apparatus prior to the NORT for 5 min, and then two trials in the NORT, an acquisition trial (trial 1) and a retention trial (trial 2). These two trials are separated by a inter-trial interval of 15 min (short-term) and 24 hours (long-term). Each dam is placed in an arena and allowed to investigate two identical objects for 5 min. After the retention period, trial 2 is completed where dam is presented with a familiar object (one of the objects from trial 1) and a novel object (not present in trial 1). It was cleaned with 70 % ethanol before the next dam was put on the apparatus. Behaviours scored during the NORT include: discrimination Index which is the time spent between the novel and familiar objects (Shimoda *et al*., 2021).

$$\text{Discrimination Index} = \frac{\text{Novel object exploration time} - \text{Familiar object exploration time}}{\text{Novel object exploration time} + \text{Familiar object exploration time}}$$

Habituation Index: Total time spent in exploring the two objects during the familiarization phase and the test phase (Antunes and Biala, 2012).

T-maze spontaneous alternation test: The T-maze is an elevated or enclosed apparatus in the form of a 'T' placed horizontally. It is made of wood painted black and consist of three arms, the starting arm which is the base of the T-maze measuring 50 x 16 cm and two goal arms (left and right) measuring 50 x 10 cm with a wooden door cut to fit at the entrance, which give animal two environment to explore. The wall height of the enclosed T-maze is 30 cm. On PPD 15, each dam was placed on the maze and allowed

to explore the maze for 5 min. This maze was used for 2-trials and the maze was cleaned with 70% ethanol before the next rat was put on the apparatus. Prior to Trial 1, the left and right arm doors of the maze was open and the rat was placed at the base of the Maze, and a stopwatch was used to record the time it took the animal to explore the goal arms. When all four paws of dam enter one arm, the arm door is closed for 2 min and arm entry is recorded as right (R) or left (L). Trial 2 was done after 15 min, the same procedure was followed as Trial 1.

Spontaneous alternation behaviour (SAB) is based on the fact that rodents prefer to visit the less recently visited arm, thus implicating that it will need to recall which was the last arm visited. The percentage alternation was calculated as reported by Sivakumar *et al*., (2017).

Percentage alternation:

$$\frac{\text{The number of correct choices X 100}}{\text{Total sets performed}}$$

Assessment of social behaviour

Nesting: On PPD 5 cotton wool was provided as nesting material. Five gram (5g) of the nesting material was placed in the home cage and examined after 24 hours for nest building behaviour. Nest scoring include: (a) Nestlet not noticeably touched (90% or more intact), (b) Nestlet partially torn (50-90% intact), (c) Less than 50% of nestlet remains intact, but not gathered into a nest site but spread throughout cage, (d) More than 90% of the nestlet is torn into a flat nest (e) More than 90% of the nestlet is torn, nest is fairly even (Deacon, 2006).

Assessment of pain sensation

Hot Plate Test: On PPD 19, each dam was dropped gently on the hot-plate (Hotplate analgesia meter Columbus instruments OHIO-43204 USA) set at 55±1°C. The reaction time was recorded using a stopwatch as the interval between placement of the animals on the hot plate and the first time it licked its fore-paws (Yam *et al*., 2020).

Tail flick test:: This experiment was conducted according to the modified method adopted by Sanchez-Mateo *et al*., (2006) using hot water bath. On PPD 17, the terminal 2 cm of the rat tail in each group was immersed in hot water contained in a 500 ml beaker maintained at 55±1°C. A thermometer was placed inside the water to monitor the temperature (Sanchez-Mateo *et al*., 2006). The time in seconds to clearly withdraw the tail out of the water was taken as the reaction time, with a cut-off time of immersion at 10 sec (Sudipta *et al*., 2013).

Statistical Analysis:

Data were expressed as Mean ± Standard Error of Mean (SEM). Sample size for each treatment group is stated in respective table or figures. Data were first examined for normality using Shapiro-Wilk normality test, with $p>0.05$ the null hypothesis was accepted signifying normal distribution of the data. Normally distributed data were analyzed with independent Student's t-test for comparison between means of the two groups. A difference between means was considered significant at $p < 0.05$. The statistical software used include SPSS version 20 and graphpad prism version 8.

RESULTS

Acute toxicity study: The median lethal dose (LD50) value of the aqueous leaf extract of JT leaves in rat was found to be greater than 5000 mg/kg body weight, orally. There was no toxicity and mortality recorded even at the highest dose of 5000mg/kg.

Neurobehavioural observation

Antidepressant-like activity of JT in dams: In FST, The mean latency to immobility for control and JT treated dams are 70.61 ± 7.59 and 103.88 ± 10.31 respectively. Latency to immobility was significantly ($P<0.05$) higher in the JT group when compared to the control (Figure 1).

The mean duration of immobility for control and JT treated dams are 64.89 ± 7.23 and 45.61 ± 4.99 respectively. Duration of immobility was significantly ($P<0.05$) reduced in the JT group when compared to the control (Figure 2).

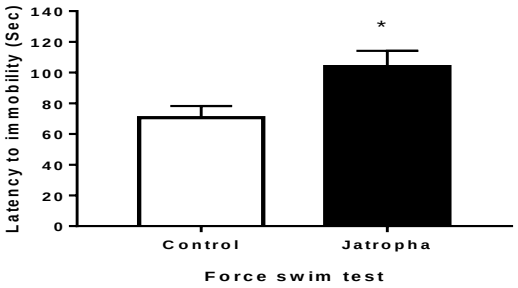

Figure. 1:
Comparison of latency to immobility in FST between experimental groups
Values are mean ± SEM, n= control (7); Jatropha (9)
*p<0.05 vs control

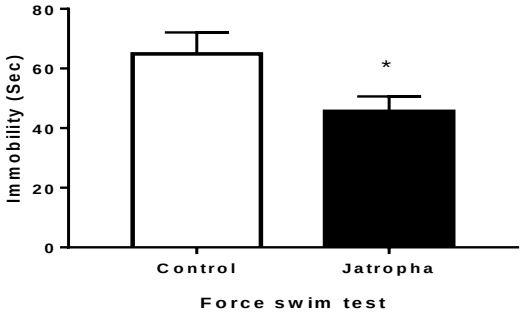

Figure. 2:
Comparison of Immobility in FST between experimental groups
Values are mean ± SEM, n= control (7); Jatropha (9) *p<0.05 vs control

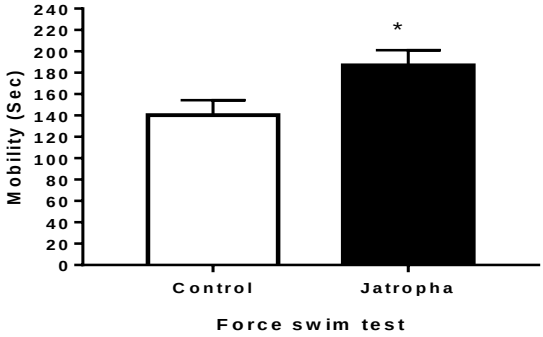

Figure. 3:
Comparison of mobility in FST between experimental groups Values are mean ± SEM, n= control (7); Jatropha (9)
*p<0.05 vs control

The mean duration of mobility for control and JT treated dams are 140.22 ± 14.13 and 186.80 ± 14.31 respectively. Duration of mobility was significantly (P<0.05) higher in the JT group when compared to the control (Figure 3).
The mean frequency of defecation for control and JT treated dams are 7.29 ± 0.81 and 3.22 ± 0.55 respectively. Frequency of defecation was significantly (P<0.01) higher in the control when compared to the JT group (Figure 4). In TST, the mean latency to immobility for control and JT treated dams are 48.47 ± 6.29 and 75.84 ± 7.83 respectively. Latency to immobility was significantly (P<0.05) higher in the JT group when compared to the control (Figure 5). The mean duration of immobility for control and JT treated dams are 97.03 ± 6.81 and 34.09 ± 2.59 respectively. Duration of immobility was significantly (P<0.001) higher in the control when compared to JT group (Figure 6). The mean duration of mobility for control and JT treated dams are 154.93 ± 8.96 and 190.62 ± 9.24 respectively. Duration of mobility was significantly (P<0.05) higher in the control when compared to JT group (Figure 7).

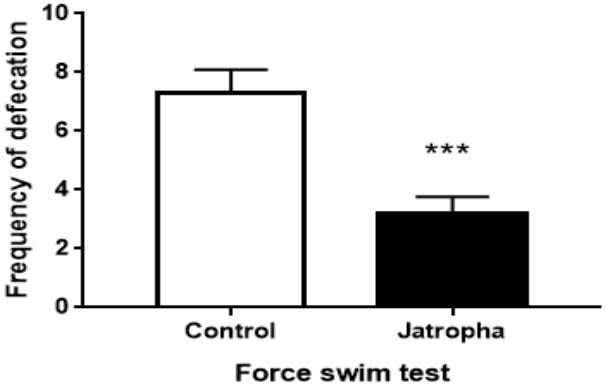

Figure. 4:
Comparison of frequency of defecation in FST between experimental groups
Values are mean ± SEM, n= control (7); Jatropha (9)
***p<0.001 vs control

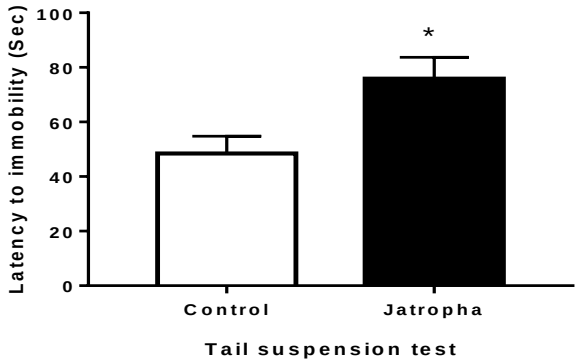

Figure. 5:
Comparison of latency to immobility in TST between experimentalgroups
Values are mean ± SEM, n= control (7); Jatropha (9)
*p<0.05 vs control.

Anxiolytic-like activity of JT in dams: Table 1 shows the effects aqueous leaf extract of JT in dams subjected to anxiolytic-like activity in EPM. Results obtained indicate that administration of JT caused a significant (P<0.05) decrease in the frequency of head dip, urination, defecation, grooming and it duration compared to control. The frequency of SAP and close arm frequency were significantly (P<0.01) lower in the JT group compared to control. The close arm duration was significantly (P<0.001) lower in the JT group compared to control. The frequency of entry into the open arm (P<0.001) and open arm duration (P<0.01) were significantly higher in the JT group compared to control.

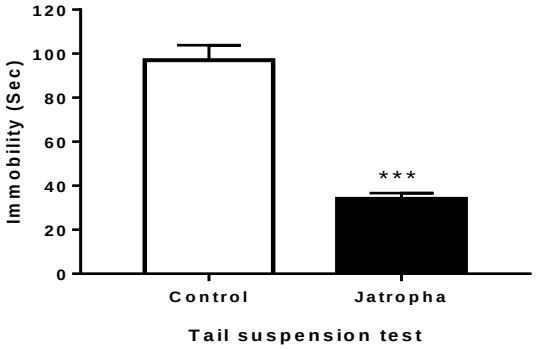

Figure. 6:
Comparison of immobility in TST between experimental groups
Values are mean ± SEM, n= control (7); Jatropha (9)
***p<0.001 vs control

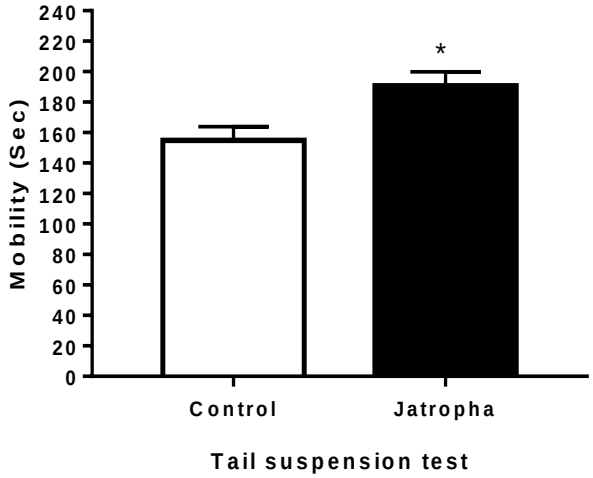

Figure. 7:
Comparison of mobility in TST between experimental groups
Values are mean ± SEM, n= control (7); Jatropha (9)
*p<0.05 vs control

Table 1:
Comparison of EPM activities in JT dams

Parameters	Control	Jatropha
Head dipping	7.86 ± 1.5	3.44 ± 1.06*
Rearing	11.29 ± 1.41	17.56 ± 2.06*
Stretch attend posture	7.43 ± 0.97	3.33 ± 0.58**
Grooming frequency	3.57 ± 0.97	1.00 ± 0.44*
Grooming duration	45.72 ± 15.91	6.01 ± 2.47*
Open arm frequency	0.86 ± 0.26	2.78 ± 0.32***
Open arm duration	15.50 ± 7.31	162.58 ± 30.73**
Close arm frequency	2.00 ± 0.22	1.22 ± 0.15**
Close arm duration	284.46 ± 7.31	99.72 ± 31.36***
Urination	3.00 ± 0.82	0.33 ± 0.24*
Defecation	2.29 ± 0.57	0.78 ± 0.22*

Values are Mean ± SEM, n= control (7); Jatropha (9) * P<0.05, ** P<0.01, *** P<0.001 significantly different compared to control.

Table 2 shows the summary of behaviours scored in the LDB test following treatment with the aqueous leaf-extract of JT. The frequency of line crossing, rearing, transitioning in the light and dark chamber, as well as duration in the light box were significantly (P<0.05) higher in the JT group compared to control. The frequency of SAP and duration in the dark box were significantly (P<0.05) lower in the JT group compared to control. The duration of grooming was significantly (P<0.01) lower in the JT group compared to control.

Table 2:
LDB test of dams treated with JT

Parameters	Control	Jatropha
Line crossing	57.29 ± 7.10	74.67 ± 3.82*
Rearing	18.86 ± 4.07	30.0 ± 1.82*
Transition frequency	6.57 ± 1.2	10.89 ± 1.02*
SAP	2.71 ± 0.29	1.44 ± 0.41*
Grooming	38.34 ± 7.07	14.57 ± 3.25**
Light box duration	60.30 ± 13.07	150.16 ± 32.90*
Dark box duration	215.62 ± 19.38	106.59 ± 35.85*
Urination	4.86 ± 0.74	2.00 ± 0.76*
Defecation	4.57 ± 0.81	2.00 ± 0.65*

Values are Mean ± SEM, n= control (7); Jatropha (9) * P<0.05, ** P<0.01 significantly different compared to control.

Table 3:
OFT of dams treated with JT

Parameters	Control	Jatropha
Line crossing	63.14 ± 8.05	108.44 ± 11.50**
Rearing	21.57 ± 3.75	40.89 ± 4.51**
SAP	3.00 ± 1.02	0.44 ± 0.29*
Grooming frequency	3.71 ± 0.89	3.44 ± 0.94^{NS}
Grooming duration	32.23 ± 9.41	25.72 ± 7.46^{NS}
Freezing frequency	4.71 ± 0.64	2.11 ± 0.56**
Freezing duration	72.73 ± 10.74	32.24 ± 12.92*
CSE frequency	1.43 ± 0.48	4.33 ± 1.12*
CSE duration	9.90 ± 4.73	45.91 ± 13.19*
Urination	4.86 ± 1.18	2.00 ± 0.62*
Defecation	3.71 ± 0.78	1.56 ± 0.50*

Values are Mean ± SEM, n= control (7); Jatropha (9) * P<0.05, ** P<0.01 significantly different compared to control. NS= Not significant

Locomotor activity and emotionality using open field maze: The frequency of line crossing, rearing (P<0.01), CSE and duration were significantly (P<0.05) higher in the JT group compared to control, while the frequency of SAP, urination, defecation and duration of freezing were significantly (P<0.05) lower in the JT group compared to control. The freezing frequency was significantly (P<0.01) lower in the JT group compared to control, as shown in Table 3.

Social behaviour: Observations of nest building for control and JT treated group are: 2.71 ± 0.47 and 4.11 ± 0.26 respectively. Nest building increased significantly (P<0.05) in the JT group compared to the control (Figure 8).

Cognitive function: Results for NORT showed the mean index of habituation for short term memory (STM) of control and treated dams as 5.63 ± 2.85 and 3.11 ± 3.26 respectively. There was no significant differences among group (Figure 9).

The mean index of habituation for long term memory (LTM) of control and treated dams are -2.28 ± 2.85 and 1.06 ± 5.86 respectively. There was no significant differences among group (Figure 10). The mean index of discrimination for STM of control and treated dams are -0.005 ± 0.12 and 0.26 ± 0.17 respectively. There was no significant differences among group (Figure 11). The mean index of discrimination for LTM of control and treated dams are 0.02 ± 0.21 and 0.19 ± 0.21 respectively. There was no significant difference among group (Figure 12).

T -Maze test results for STM showed the percentage alternation for control and treated dams as 42.86 ± 20.20 and 77.78 ± 14.70 respectively. There was no significant difference among group (Figure 13).

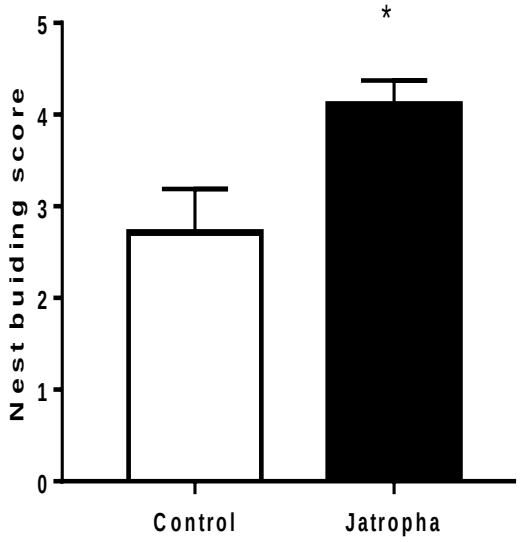

Figure. 8:
Comparison of nest building score between experimental group
Values are mean ± SEM, n= control (7); Jatropha
*p<0.05 vs control

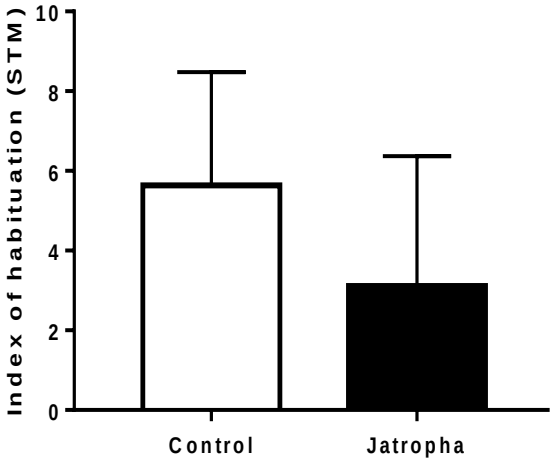

Figure 9:
Comparison of Index of habituation (STM) in NORT between experimental groups.
Values are mean ± SEM, n= control (7); Jatropha (9)

Pain sensation: The mean retraction time of HPT for control and JT treated dams are 11.03 ± 1.40 and 11.79 ±

2.20 respectively. There was no significant differences between the groups (Figure 14).

The mean retraction time of TFT test for control and treated dams are 6.19 ± 0.83 and 9.78 ± 0.82 respectively. There was significant ($P<0.01$) increase in the retraction time of the JT dams compared to the control (Figure 15).

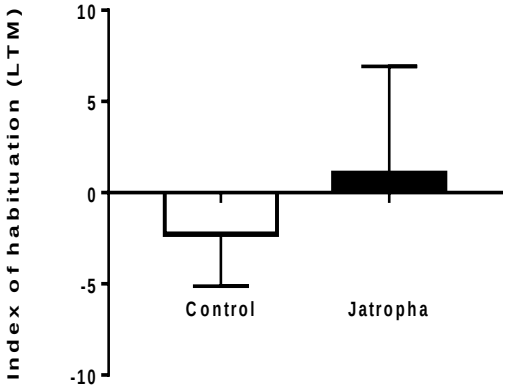

Figure 10:
Comparison of Index of habituation (LTM) in NORT between experimental groups.
Values are mean ± SEM, n= control (7); Jatropha (9)

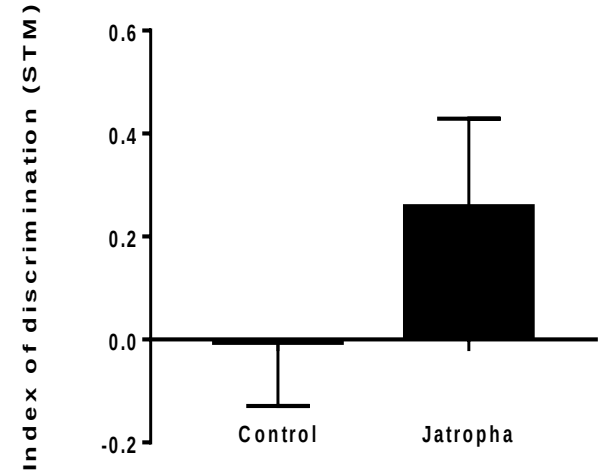

Figure 11:
Comparison of Index of discrimination (STM) in NORT between experimental groups.
Values are mean ± SEM, n= control (7); Jatropha (9)

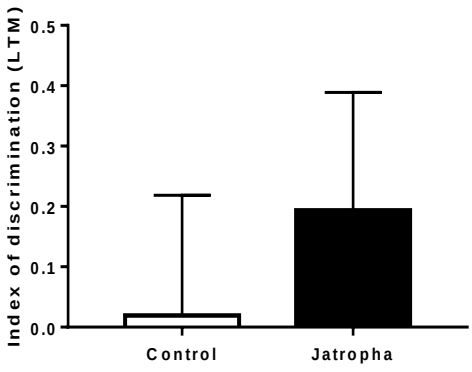

Figure 12:
Comparison of Index of discrimination (LTM) in NORT between experimental groups.
Values are mean ± SEM, n= control (7); Jatropha (9)

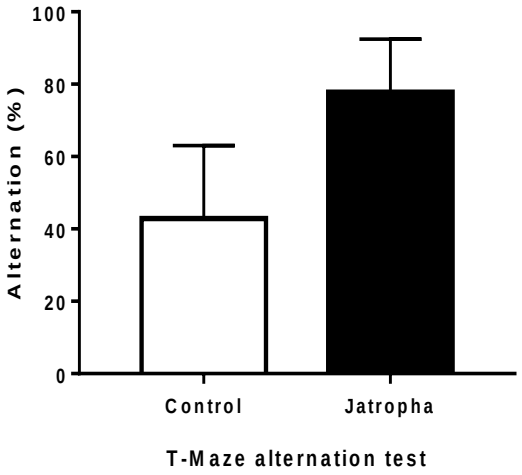

Figure 13:
Comparison of percentage alternation between experimental groups.
Values are mean ± SEM, n= control (7); Jatropha (9)

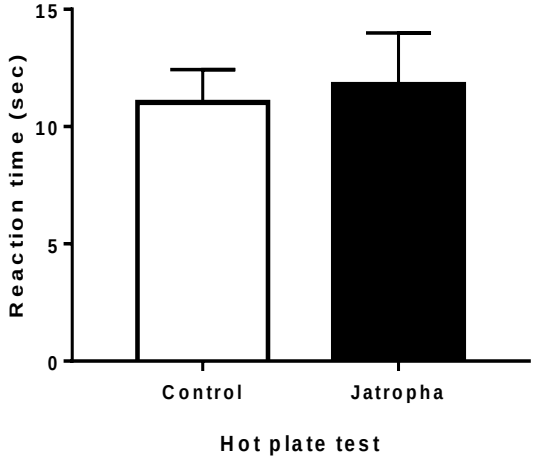

Figure 14:
Comparison of reaction time in HPT between experimental groups.
Values are mean ± SEM, n= control (7); Jatropha (9)

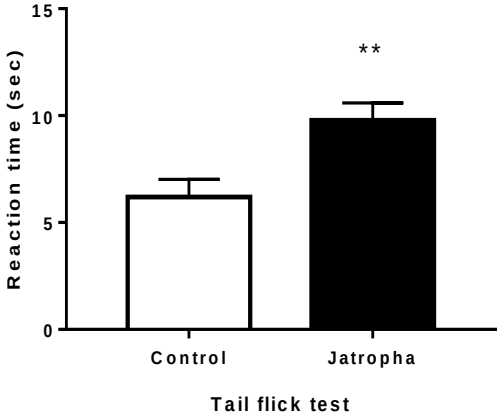

Figure 15:

Comparison of reaction time in TFT between experimental groups
Values are mean ± SEM, n= control (7); Jatropha (9)
**- Significant at p< 0.01 versus control

DISCUSSION

The occurrence of postpartum disorders after childbirth is well known during postpartum, but the treatment options for these symptoms during this special period remain limited (Zhang *et al*., 2021). In the present study, we reported that gestational administration of aqueous leaf extract of JT significantly contributed to the alleviation of postpartum-like behaviour in rats. Antidepressant-like profile was observed in JT treated dams; this is evidenced by the increased duration of latency to immobility and mobility, as well as decrease duration of immobility in both the FST and TST. Immobility has been considered to reflect behavioural despair similar to that seen in human depression, and antidepressant medications are able to reduce the immobility time in rodents (Vanzella *et al*., 2012), whereas latency to the first immobility is a measure of active stress coping (Everton *et al*., 2018). It is very likely that JT contains active principles that could lessen postpartum-like depression. Some studies have reported similar antidepressant-like potential of Hypericum perforatum (Vieira *et al*., 2013), methanol root bark extract of Securinega virosa (Shehu *et al*., 2017) and folic acid (Zhang *et al*., 2021) at postpartum in rodent.

The results of EPM and LDB showed that JT-treated dams exhibited a preference for the open arm of the EPM and the light arm of the LDB. This behaviour is considered an anxiolytic-like profile (Habr *et al*., 2011, La-Vu *et al*., 2020). Habr *et al*., (2011) reported that dams exhibited decreased time spent in the open arms of the EPM. Nic Dhonnchadha *et al*., (2003) reported that dams with less anxiety-like level tend to venture more into the open arm of the EPM and the light arm of the LDB. According to Kraeuter *et al*., (2019), the avoidance of the open arm in EPM is an indicative of anxiety-like activity, while less anxiety-like animals tend to explore the environment more. The JT-treated dams made higher transitions between the brightly illuminated area and the dark arm of the LDB. Miller *et al*., (2011) reported that rodents exhibiting higher anxiety-like levels make fewer transitions between chambers in the LDB. Similarly, anxiolytic-like potential of Hypericum perforatum (Vieira *et al*., 2013) and folic acid (Zhang *et al*., 2021) at postpartum in rodent was reported.
Studies have conclusively linked results from OFT with other measures of anxiety in rodent models (Seibenhener and Wooten 2015)). Miller *et al*., (2011) reported that convergence of results among multiple behavioural paradigms would increase assurance about the most relevant influences on postpartum anxiety-like behaviour. Studies comparing locomotor activities between dams and virgin rats in OFT reported that dams exhibited decreased locomotion (Silva *et al*., 1997), while Habr *et al*., (2011) observed decreased locomotion and rearing frequency in dams suggesting increased anxiety-like and emotionality behaviour. The aqueous leaf extract of JT increased locomotor activity by increasing the frequency of line crossings and rearing in LDB and OFT as well as rearing frequency in EPM. Locomotor activity is considered as an index of alertness (Yadav *et al*., 2008). The high frequency of such behaviours indicates increased locomotion and exploration, and refers to a lower anxiety-like level (Zimcikovaa *et al*., 2017). Increase in line crosses, rearing and transition frequency in both LDB and OFT observed in JT is an indication of CNS stimulant properties (Harquin *et al*., 2012).

Several well-known antidepressants was reported to decrease locomotor activity (Shehu *et al*., 2017). Contrary to this study, the aqueous leaf extract of JT not only reduces postpartum-like depression, it as well increase locomotor activity. La-Vu *et al*., (2020) reported that rodents in all paradigms demonstrating an increase in SAP, defecation, and urination indicate greater anxiety-like behaviour. Results from EPM, OFT and LDB showed a reduction in these behaviours, FST also showed reduced defecation frequency signifying that JT was able to reduce anxiety-like behaviour. SAP is a good identifier for exploratory and anxiety-like conflict behaviour in rodent and can be used to evaluate the effects of medications at reducing these internal conflicts (Holly *et al*., 2016). JT was effective in reducing this conflict in dams. Defecation is a parameter that indicates an increase in emotionality (Habr *et al*., 2011), JT improved emotionality by reducing the frequency of defecation.

Holmes *et al*., (2003) reported that increase grooming and head dip in EPM and grooming in LDB specify increased anxiety-like levels. JT treated dams reduced these behaviours in EPM and LDB but did not reduce grooming in OFT. Silva *et al*., (1997) reported that an increase in anxiety may lead to a freezing that ultimately leads to a reduction in OFT locomotion activity. In this study the frequency and duration of freezing was significantly reduced. This may be due to the effect of JT in lessening postpartum-like anxiety thereby increasing locomotor activity.

In this study, JT treated dams showed increased CSE and the duration of time spent in the central square. Brown *et al*., (2004) reported that high frequency/duration of these behaviours indicates high exploratory behaviour and low anxiety-like levels.

The percentage alternations was not significantly different in the JT group compared to control. The result index of discrimination in NORT can vary between +1 and -1, where a positive score indicates more time spent with the novel object, a negative score indicates more time spent with the familiar object, and a zero score indicates a null preference (Burke *et al*., 2010). Index of discrimination for STM in NORT showed a positive score in JT group compared to negative in control suggesting better cognitive function. Index of discrimination for LTM was not significant in JT group compared to control. Investigation of STM during index of habituation did not differ significantly, while LTM showed a positive score in JT group compared to control. This index and T-Maze percentage alternation however did not differ between control and JT group indicating no change in retention and hence memory.

Nest building is a common behavior in rodents, at the same time it is a kind of maternal behavior and a social behavior, associated with maintaining body temperature. The treated dam had a higher nesting score which means they showed higher interaction and building of nest.

The index of pain is measured via the escape behavior, withdrawal reflexes, licking behaviors and vocalization of these rodents which becomes the substitute for human pain

experiences in animal models (Negus *et al*., 2006). JT altered latency to painful thermal stimulus TFT. The extension of the latency time in the TFT is related to the central analgesic effect of administered drugs (Negus *et al*., 2006). Thus, this study suggest that the extracts may modulate central nociceptive pathways.

In conclusion, the result shows that aqueous leaf extract of JT has a therapeutic effect that reduces postpartum-like depression and anxiety, and possess CNS stimulant effect with improve locomotor activity in dams. This may be developed as a preventive and adjuvant therapeutic option for pregnant women.

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